



SEQUENCE LISTING

<110> Human Genome Science, Inc.  
Principia Pharmaceutical Corporation

<120> Albumin Fusion Proteins

<130> PF543

<140> 09/833,117

<141> 2001-04-12

<150> 60/229,358

<151> 2000-04-12

<150> 60/256,931

<151> 2000-12-21

<150> 60/199,384

<151> 2000-04-25

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<170> PatentIn Ver. 2.1

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<223> invertase leader sequence

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<223> first 5 amino acids of mature human serum albumin

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 gaa aat ttc aaa gcc ttg gtg ttg att gcc ttt gct cag tat ctt cag 96  
 Glu Asn Phe Lys Ala Leu Val Leu Ile Ala Phe Ala Gln Tyr Leu Gln  
 20 25 30  
 cag tgt cca ttt gaa gat cat gta aaa tta gtg aat gaa gta act gaa 144  
 Gln Cys Pro Phe Glu Asp His Val Lys Leu Val Asn Glu Val Thr Glu  
 35 40 45  
 ttt gca aaa aca tgt gtt gct gat gag tca gct gaa aat tgt gac aaa 192  
 Phe Ala Lys Thr Cys Val Ala Asp Glu Ser Ala Glu Asn Cys Asp Lys  
 50 55 60  
 tca ctt cat acc ctt ttt gga gac aaa tta tgc aca gtt gca act ctt 240  
 Ser Leu His Thr Leu Phe Gly Asp Lys Leu Cys Thr Val Ala Thr Leu  
 65 70 75 80  
 cgt gaa acc tat ggt gaa atg gct gac tgc tgt gca aaa caa gaa cct 288  
 Arg Glu Thr Tyr Gly Glu Met Ala Asp Cys Cys Ala Lys Gln Glu Pro  
 85 90 95

gag aga aat gaa tgc ttc ttg caa cac aaa gat gac aac cca aac ctc	336
Glu Arg Asn Glu Cys Phe Leu Gln His Lys Asp Asp Asn Pro Asn Leu	
100 105 110	
ccc cga ttg gtg aga cca gag gtt gat gtg atg tgc act gct ttt cat	384
Pro Arg Leu Val Arg Pro Glu Val Asp Val Met Cys Thr Ala Phe His	
115 120 125	
gac aat gaa gag aca ttt ttg aaa aaa tac tta tat gaa att gcc aga	432
Asp Asn Glu Glu Thr Phe Leu Lys Lys Tyr Leu Tyr Glu Ile Ala Arg	
130 135 140	
aga cat cct tac ttt tat gcc ccg gaa ctc ctt ttc ttt gct aaa agg	480
Arg His Pro Tyr Phe Tyr Ala Pro Glu Leu Leu Phe Phe Ala Lys Arg	
145 150 155 160	
tat aaa gct gct ttt aca gaa tgt tgc caa gct gct gat aaa gct gcc	528
Tyr Lys Ala Ala Phe Thr Glu Cys Cys Gln Ala Ala Asp Lys Ala Ala	
165 170 175	
tgc ctg ttg cca aag ctc gat gaa ctt cgg gat gaa ggg aag gct tcg	576
Cys Leu Leu Pro Lys Leu Asp Glu Leu Arg Asp Glu Gly Lys Ala Ser	
180 185 190	
tct gcc aaa cag aga ctc aaa tgt gcc agt ctc caa aaa ttt gga gaa	624
Ser Ala Lys Gln Arg Leu Lys Cys Ala Ser Leu Gln Lys Phe Gly Glu	
195 200 205	
aga gct ttc aaa gca tgg gca gtg gct cgc ctg agc cag aga ttt ccc	672
Arg Ala Phe Lys Ala Trp Ala Val Ala Arg Leu Ser Gln Arg Phe Pro	
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aaa gct gag ttt gca gaa gtt tcc aag tta gtg aca gat ctt acc aaa	720
Lys Ala Glu Phe Ala Glu Val Ser Lys Leu Val Thr Asp Leu Thr Lys	
225 230 235 240	
gtc cac acg gaa tgc tgc cat gga gat ctg ctt gaa tgt gct gat gac	768
Val His Thr Glu Cys Cys His Gly Asp Leu Leu Glu Cys Ala Asp Asp	
245 250 255	
agg gcg gac ctt gcc aag tat atc tgt gaa aat cag gat tcg atc tcc	816
Arg Ala Asp Leu Ala Lys Tyr Ile Cys Glu Asn Gln Asp Ser Ile Ser	
260 265 270	
agt aaa ctg aag gaa tgc tgt gaa aaa cct ctg ttg gaa aaa tcc cac	864
Ser Lys Leu Lys Glu Cys Cys Glu Lys Pro Leu Leu Glu Lys Ser His	
275 280 285	

tgc att gcc gaa gtg gaa aat gat gag atg cct gct gac ttg cct tca	912
Cys Ile Ala Glu Val Glu Asn Asp Glu Met Pro Ala Asp Leu Pro Ser	
290 295 300	
tta gct gct gat ttt gtt gaa agt aag gat gtt tgc aaa aac tat gct	960
Leu Ala Ala Asp Phe Val Glu Ser Lys Asp Val Cys Lys Asn Tyr Ala	
305 310 315 320	
gag gca aag gat gtc ttc ctg ggc atg ttt ttg tat gaa tat gca aga	1008
Glu Ala Lys Asp Val Phe Leu Gly Met Phe Leu Tyr Glu Tyr Ala Arg	
325 330 335	
agg cat cct gat tac tct gtc gtg ctg ctg ctg aga ctt gcc aag aca	1056
Arg His Pro Asp Tyr Ser Val Val Leu Leu Leu Arg Leu Ala Lys Thr	
340 345 350	
tat gaa acc act cta gag aag tgc tgt gcc gct gca gat cct cat gaa	1104
Tyr Glu Thr Thr Leu Glu Lys Cys Cys Ala Ala Ala Asp Pro His Glu	
355 360 365	
tgc tat gcc aaa gtg ttc gat gaa ttt aaa cct ctt gtg gaa gag cct	1152
Cys Tyr Ala Lys Val Phe Asp Glu Phe Lys Pro Leu Val Glu Glu Pro	
370 375 380	
cag aat tta atc aaa caa aac tgt gag ctt ttt gag cag ctt gga gag	1200
Gln Asn Leu Ile Lys Gln Asn Cys Glu Leu Phe Glu Gln Leu Gly Glu	
385 390 395 400	
tac aaa ttc cag aat gcg cta tta gtt cgt tac acc aag aaa gta ccc	1248
Tyr Lys Phe Gln Asn Ala Leu Leu Val Arg Tyr Thr Lys Lys Val Pro	
405 410 415	
caa gtg tca act cca act ctt gta gag gtc tca aga aac cta gga aaa	1296
Gln Val Ser Thr Pro Thr Leu Val Glu Val Ser Arg Asn Leu Gly Lys	
420 425 430	
gtg ggc agc aaa tgt tgt aaa cat cct gaa gca aaa aga atg ccc tgt	1344
Val Gly Ser Lys Cys Cys Lys His Pro Glu Ala Lys Arg Met Pro Cys	
435 440 445	
gca gaa gac tat cta tcc gtg gtc ctg aac cag tta tgt gtg ttg cat	1392
Ala Glu Asp Tyr Leu Ser Val Val Leu Asn Gln Leu Cys Val Leu His	
450 455 460	
gag aaa acg cca gta agt gac aga gtc aca aaa tgc tgc aca gag tcc	1440
Glu Lys Thr Pro Val Ser Asp Arg Val Thr Lys Cys Cys Thr Glu Ser	
465 470 475 480	
ttg gtg aac agg cga cca tgc ttt tca gct ctg gaa gtc gat gaa aca	1488



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Tyr	Val	Pro	Lys	Glu	Phe	Asn	Ala	Glu	Thr	Phe	Thr	Phe	His	Ala	Asp	
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ata	tgc	aca	ctt	tct	gag	aag	gag	aga	caa	atc	aag	aaa	caa	act	gca	1584
Ile	Cys	Thr	Leu	Ser	Glu	Lys	Glu	Arg	Gln	Ile	Lys	Lys	Gln	Thr	Ala	
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ctt	gtt	gag	ctt	gtg	aaa	cac	aag	ccc	aag	gca	aca	aaa	gag	caa	ctg	1632
Leu	Val	Glu	Leu	Val	Lys	His	Lys	Pro	Lys	Ala	Thr	Lys	Glu	Gln	Leu	
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Lys	Ala	Val	Met	Asp	Asp	Phe	Ala	Ala	Phe	Val	Glu	Lys	Cys	Cys	Lys	
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Ala	Asp	Asp	Lys	Glu	Thr	Cys	Phe	Ala	Glu	Glu	Gly	Lys	Lys	Leu	Val	
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gct	gca	agt	caa	gct	gcc	tta	ggc	tta	taacatctac attttaaagc atctcag							1782
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Gln	Cys	Pro	Phe	Glu	Asp	His	Val	Lys	Leu	Val	Asn	Glu	Val	Thr	Glu
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Arg	Glu	Thr	Tyr	Gly	Glu	Met	Ala	Asp	Cys	Cys	Ala	Lys	Gln	Glu	Pro	85	90	95
Glu	Arg	Asn	Glu	Cys	Phe	Leu	Gln	His	Lys	Asp	Asp	Asn	Pro	Asn	Leu	100	105	110
Pro	Arg	Leu	Val	Arg	Pro	Glu	Val	Asp	Val	Met	Cys	Thr	Ala	Phe	His	115	120	125
Asp	Asn	Glu	Glu	Thr	Phe	Leu	Lys	Lys	Tyr	Leu	Tyr	Glu	Ile	Ala	Arg	130	135	140
Arg	His	Pro	Tyr	Phe	Tyr	Ala	Pro	Glu	Leu	Leu	Phe	Phe	Ala	Lys	Arg	145	150	155
Tyr	Lys	Ala	Ala	Phe	Thr	Glu	Cys	Cys	Gln	Ala	Ala	Asp	Lys	Ala	Ala	165	170	175
Cys	Leu	Leu	Pro	Lys	Leu	Asp	Glu	Leu	Arg	Asp	Glu	Gly	Lys	Ala	Ser	180	185	190
Ser	Ala	Lys	Gln	Arg	Leu	Lys	Cys	Ala	Ser	Leu	Gln	Lys	Phe	Gly	Glu	195	200	205
Arg	Ala	Phe	Lys	Ala	Trp	Ala	Val	Ala	Arg	Leu	Ser	Gln	Arg	Phe	Pro	210	215	220
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Val	His	Thr	Glu	Cys	Cys	His	Gly	Asp	Leu	Leu	Glu	Cys	Ala	Asp	Asp	245	250	255
Arg	Ala	Asp	Leu	Ala	Lys	Tyr	Ile	Cys	Glu	Asn	Gln	Asp	Ser	Ile	Ser	260	265	270
Ser	Lys	Leu	Lys	Glu	Cys	Cys	Glu	Lys	Pro	Leu	Leu	Glu	Lys	Ser	His	275	280	285
Cys	Ile	Ala	Glu	Val	Glu	Asn	Asp	Glu	Met	Pro	Ala	Asp	Leu	Pro	Ser	290	295	300
Leu	Ala	Ala	Asp	Phe	Val	Glu	Ser	Lys	Asp	Val	Cys	Lys	Asn	Tyr	Ala	305	310	315
Glu	Ala	Lys	Asp	Val	Phe	Leu	Gly	Met	Phe	Leu	Tyr	Glu	Tyr	Ala	Arg	325	330	335
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of the Therapeutic Protein

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51

<210> 27  
<211> 33  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> forward primer useful for generation of albumin fusion protein in which the albumin moiety is c-terminal of the Therapeutic Protein

<220>  
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<220>

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<400> 27  
aggagcgtcg acaaaagann nnnnnnnnnn nnn

33

<210> 28  
<211> 52  
<212> DNA  
<213> Artificial Sequence

<220>  
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<223> reverse primer useful for generation of albumin  
fusion protein in which the albumin moiety is c-terminal of  
the Therapeutic Protein

<220>  
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<222> (38)  
<223> n equals a,t,g, or c

<220>  
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<220>  
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<400> 28  
ctttaaatcg atgagcaacc tcactcttgt gtgcacnnn nnnnnnnnnn nn

52

<210> 29  
 <211> 24  
 <212> PRT  
 <213> Artificial Sequence  
  
 <220>  
 <221> signal  
 <223> signal peptide of natural human serum albumin protein  
  
 <400> 29  
 Met Lys Trp Val Ser Phe Ile Ser Leu Leu Phe Leu Phe Ser Ser Ala  
     1                    5                    10                    15  
  
 Tyr Ser Arg Ser Leu Asp Lys Arg  
                     20  
  
 <210> 30  
 <211> 114  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <221> primer\_bind  
 <223> forward primer useful for generation of PC4:HSA  
 albumin fusion VECTOR  
  
 <220>  
 <221> misc\_feature  
 <222> (5)..(10)  
 <223> BamHI restriction site  
  
 <220>  
 <221> misc\_feature  
 <222> (11)..(16)  
 <223> Hind III restriction site  
  
 <220>  
 <221> misc\_feature  
 <222> (17)..(27)  
 <223> Kozak sequence  
  
 <220>  
 <221> misc\_feature  
 <222> (25)..(97)  
 <223> cds natural signal sequence of human serum albumin  
  
 <220>  
 <221> misc\_feature  
 <222> (75)..(81)

<223> XhoI restriction site

<220>  
 <221> misc\_feature  
 <222> (98)..(114)  
 <223> cds first six amino acids of human serum albumin

<400> 30  
 tcagggatcc aagcttccgc caccatgaag tgggtaacct ttatttcctc tctttttctc 60  
 tttagctcgg cttactcgag ggggtgtgttt cgtcgagatg cacacaagag tgag 114

<210> 31  
 <211> 43  
 <212> DNA  
 <213> Artificial Sequence

<220>  
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 <223> reverse primer useful for generation of  
 PC4:HSA albumin fusion VECTOR

<220>  
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 <222> (6)..(11)  
 <223> Asp718 restriction site

<220>  
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 <222> (12)..(17)  
 <223> EcoRI restriction site

<220>  
 <221> misc\_feature  
 <222> (15)..(17)  
 <223> reverse complement of stop codon

<220>  
 <221> misc\_feature  
 <222> (18)..(25)  
 <223> AscI restriction site

<220>  
 <221> misc\_feature  
 <222> (18)..(43)  
 <223> reverse complement of DNA sequence encoding last 9 amino acids

<400> 31

gcagcggtac cgaattcggc ggccttata agcctaaggc agc

43

<210> 32

<211> 46

<212> DNA

<213> Artificial Sequence

<220>

<221> primer\_bind

<223> forward primer useful for inserting Therapeutic protein into pC4:HSA vector

<220>

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<222> (29)

<223> n equals a,t,g, or c

<220>

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<222> (30)

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<222> (31)

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<223> n equals a,t,g, or c

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<223> n equals a,t,g, or c

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<223> n equals a,t,g, or c

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<223> n equals a,t,g, or c

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<221> misc\_feature

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<221> misc\_feature  
<222> (46)  
<223> n equals a,t,g, or c

<400> 32  
ccgccgctcg aggggtgtgt ttcgtcgann nnnnnnnnnn nnnnnn

46

<210> 33  
<211> 55  
<212> DNA  
<213> Artificial Sequence

<220>  
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<223> reverse primer useful for inserting Therapeutic  
protein into pC4:HSA vector

<220>  
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<222> (38)  
<223> n equals a,t,g, or c

<220>  
<221> misc\_feature  
<222> (39)  
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<220>  
<221> misc\_feature  
<222> (40)  
<223> n equals a,t,g, or c

<220>  
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<223> n equals a,t,g, or c

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<222> (42)  
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<220>  
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<222> (44)  
<223> n equals a,t,g, or c

<220>  
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<223> n equals a,t,g, or c

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<220>  
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<220>  
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<223> n equals a,t,g, or c

<220>  
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<223> n equals a,t,g, or c

<220>

<221> misc\_feature  
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<223> n equals a,t,g, or c

<220>  
<221> misc\_feature  
<222> (55)  
<223> n equals a,t,g, or c

<400> 33  
agtcccatcg atgagcaacc tcactcttgt gtgcatcnnn nnnnnnnnnn nnnnn 55

<210> 34  
<211> 17  
<212> PRT  
<213> Artificial Sequence

<220>  
<221> signal  
<223> Stanniocalcin signal peptide

<400> 34  
Met Leu Gln Asn Ser Ala Val Leu Leu Leu Leu Val Ile Ser Ala Ser  
1 5 10 15

Ala

<210> 35  
<211> 22  
<212> PRT  
<213> Artificial Sequence

<220>  
<221> signal  
<223> Synthetic signal peptide

<400> 35  
Met Pro Thr Trp Ala Trp Trp Leu Phe Leu Val Leu Leu Leu Ala Leu  
1 5 10 15

Trp Ala Pro Ala Arg Gly  
20

<210> 36  
<211> 733  
<212> DNA  
<213> Homo sapiens

<400> 36

gggatccgga gcccaaattct tctgacaaaa ctcacacatg cccaccgtgc ccagcacctg	60
aattcgaggg tgcaccgtca gtcttctctt tcccccaaa acccaaggac accctcatga	120
tctcccggac tcttgaggtc acatgcgtgg tgggtggacgt aagccacgaa gaccctgagg	180
tcaagttcaa ctggtacgtg gacggcgtgg aggtgcataa tgccaagaca aagccgcggg	240
aggagcagta caacagcacg taccgtgtgg tcagcgtcct caccgtcctg caccaggact	300
ggctgaatgg caaggagtac aagtgcagg tctccaacaa agccctcca acccccatcg	360
agaaaacat ctccaaagcc aaagggcagc cccgagaacc acaggtgtac accctgcccc	420
catcccggga tgagctgacc aagaaccagg tcagcctgac ctgcctggtc aaaggcttct	480
atccaagcga catcgccgtg gagggggaga gcaatgggca gccggagaac aactacaaga	540
ccacgcctcc cgtgctggac tccgacggct ccttcttctt ctacagcaag ctcaccgtgg	600
acaagagcag gtggcagcag gggaacgtct tctcatgctc cgtgatgcat gaggctctgc	660
acaaccacta cacgcagaag agcctctccc tgtctccggg taaatgagtg cgacggccgc	720
gactctagag gat	733

<210> 37

<211> 5

<212> PRT

<213> Artificial sequence

<220>

<221> misc\_structure

<223> membrane proximal motif of class 1 cytokine receptors

<220>

<221> misc\_feature

<222> (3)

<223> Xaa equals any

<400> 37

Trp Ser Xaa Trp Ser

1

5

<210> 38

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<221> primer\_bind

<223> forward primer useful for generation of a synthetic gamma activation site (GAS) containing promoter element

<400> 38

gcgcctcgag atttccccga aatctagatt tccccgaaat gatttccccg aaatgatttc 60

cccgaaatat ctgccatctc aattag 86

<210> 39

<211> 27

<212> DNA

<213> Artificial Sequence

<220>

<221> primer\_bind

<223> reverse primer useful for generation of a synthetic gamma activation site (GAS) containing promoter element

<400> 39

gcggcaagct ttttgcaaag cctaggc 27

<210> 40

<211> 271

<212> DNA

<213> Artificial Sequence

<220>

<221> misc\_feature

<223> Synthetic GAS-SV40 promoter sequence

<400> 40

ctcgagattt ccccgaaatc tagatttccc cgaaatgatt tccccgaaat gatttccccg 60

aaatatctgc catctcaatt agtcagcaac catagtcccc cccctaactc cgcccatccc 120

gcccctaact ccgcccagtt ccgcccattc tccgcccatt ggctgactaa ttttttttat 180

ttatgcagag gccgaggccg cctcggcctc tgagctattc cagaagtagt gaggaggctt 240

ttttggaggc ctaggctttt gcaaaaagct t 271

<210> 41

<211> 32

<212> DNA

<213> Artificial Sequence

<220>  
 <221> primer\_bind  
 <223> primer useful for generation of a EGR/SEAP reporter construct  
  
 <400> 41  
 gcgctcgagg gatgacagcg atagaacccc gg 32  
  
 <210> 42  
 <211> 31  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <221> primer\_bind  
 <223> primer useful for generation of a EGR/SEAP reporter construct  
  
 <400> 42  
 gcgaagcttc gcgactcccc ggatccgcct c 31  
  
 <210> 43  
 <211> 12  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <221> misc\_binding  
 <223> NF-KB binding site  
  
 <400> 43  
 ggggactttc cc 12  
  
 <210> 44  
 <211> 73  
 <212> DNA  
 <213> Artificial Sequence  
  
 <220>  
 <221> primer\_bind  
 <223> forward primer useful for generation of a vector containing the  
 NF-KB promoter element  
  
 <400> 44  
 gcggcctcga ggggactttc ccggggactt tccggggact ttccgggact ttccatcctg 60  
 ccatctcaat tag 73  
  
 <210> 45

<211> 256  
<212> DNA  
<213> Artificial Sequence

<220>  
<221> misc\_feature  
<223> Synthetic NF-KB/SV40 promoter

<400> 45  
ctcgagggga ctttcccggg gactttccgg ggactttccg ggactttcca tctgccatct 60  
caattagtca gcaaccatag tcccgccct aactccgccc atcccgcccc taactccgcc 120  
cagttccgcc cattctccgc cccatggctg actaattttt tttatttatg cagaggccga 180  
ggccgcctcg gcctctgagc tattccagaa gtagtgagga ggcttttttg gaggcctagg 240  
cttttgcaaa aagctt 256